

**Standard Protocol for Assessing the Ecological
Condition (Quality) of Depressional Wetlands
in the Prairie Pothole Region of Iowa
Version 1.0**



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A standard protocol to assess the ecological condition of wetlands is important for multiple reasons. The main reason being that standard assessment protocols did not exist. Wetlands are inherently more difficult to assess because they are dynamic. Unlike other bodies of water, these shallow water systems change a lot from year to year and often change throughout the course of one year. Proper wetland assessment takes time and money to do and both are often at a premium. It is also important that the subsequent data and information collected from wetland assessments is done in the same way, managed the same way, and interpreted in a consistent manner. Therefore, it is crucial to sample and survey the right variables that determine wetland quality in a standardized manner so the information is comparable and useful to land managers and the public over time and across different wetland sites.

Recent wetland research and monitoring conducted in Iowa's portion of the Prairie Pothole Region has shown that multiple variables are better to use than just one variable in assessing a wetland, much like a routine physical at a doctor's office for humans. This wetland assessment protocol explains which variables are best to sample for and how to do this in order to determine the ecological health or condition of depressional, pothole wetlands in Iowa. The protocol is designed for use with a Wetland Condition Index developed for Iowa's depressional wetlands. The Wetland Condition Index (WCI) provides a standardized way of producing an assessment of a wetland from multiple variables that is easy to understand. It is important to also point out that this protocol is Version 1.0. Further refinement of wetland assessment sampling methodologies may occur in the future that simplify field work, sample processing, and/or save time, without compromising the quality of the wetland assessment data. Should this occur, it may warrant an updated version to this protocol to reflect those changes.

DESCRIPTION OF WETLAND METRICS USED IN THIS PROTOCOL

In short, a wetland metric is the measurable (quantifiable) form of a variable to survey or sample in a waterbody. The biological, physical, and chemical properties of a wetland such as aquatic plants, invertebrates, turbidity and others are all examples of variables that can be assessed (US EPA 2002). The term 'metric' will be used in this protocol to explain which variables are being measured or quantified. This wetland assessment protocol uses six metrics.

1. Total Fish Biomass
2. Chloride Concentration
3. Turbidity
4. Aquatic Plant Cover (abundance)
5. Tiger Salamander Biomass
6. Aquatic Invertebrate Taxon Richness

RATIONALE FOR WETLANDS METRICS USED IN THIS PROTOCOL

This section contains an explanation about the role of the six wetland metrics in assessing the condition of a wetland. For all sampling methods described here, these six metrics were chosen over others because they had the most statistically significant relationship in yielding information that is indicative of wetland quality. Other factors such as the ease of sampling among sites, degree of technical expertise needed by field assessment staff, sampling time, and sample costs are all considered to having a good assessment protocol.

1. **Total Fish Biomass:** Fish can directly reduce invertebrate and salamander abundance by predation (Zimmer et al. 2002; Porej and Hetherington 2005). Fish can also indirectly influence invertebrates and salamanders negatively by uprooting and consuming plants, and increasing water column concentrations of nutrients and particulate matter by excretion and disturbance of sediment (Badiou and Goldsborough 2010; Herwig et al. 2010). Larger and deeper wetlands are more likely to contain densities of fish that significantly influence the ecological condition of a wetland. The likelihood of fish successfully over-wintering increases with depth; and thus provides higher odds of successful fish reproduction and recruitment the following spring. This contributes to a higher prevalence of fish reaching maturity within the wetland system. It is important to check the area of the wetland near its outlet source; many times that is the deepest part of the wetland. Measuring the biomass is most direct method of assessing the impacts of fish to a wetland.
2. **Chloride Concentration:** It is typical to find negative relationships as salt contamination increases in a wetland. Chloride concentration is better to measure than conductivity because it is a more direct measurement. High Conductivity readings are easier to sample in the field, but the high readings may also be attributed to other ions present in the wetland, not necessarily chloride. Higher levels of chloride can directly influence invertebrate and salamanders, and possibly indirectly negatively affect them by suppressing plant growth (Mendelssohn and Batzer 2006; Griffis-Kyle 2007; Van Meter et al. 2011a). Sources of chloride contamination in a wetland can come from rural sewage effluent, industry, or road salt run-off.
3. **Turbidity:** Turbidity has been observed to increase with greater wetland area and declining depth, as wind-induced sediment suspension can increase under these conditions (Braig and Johnson 2003). Typically there is a negative correlation to be expected between turbidity and reduced aquatic plants and/or an increase in the prevalence of rough fish in a wetland. This variable is fairly quick and easy to measure while sampling in a wetland. Turbidity is a good measure of a wetland's trophic state because it strongly correlates with and is strongly influence by chlorophyll-a, nitrogen, phosphorous, and suspended solids.

4. Aquatic Plant Cover (abundance): Wetland aquatic plants provide invertebrates with habitat and food, and egg deposition sites for tiger salamanders (Euliss et al. 1999; Knutson et al. 2004). Therefore, positive relationships are likely to exist between plant abundance and diversity and the outcome variables. Using plant cover as our metric is the most beneficial way to assess the impact of plants in a wetland over other plant related metrics because it is the most direct method of abundance and can also be done by a field crew that may or may not possess strong botanical skills.
5. Tiger Salamander Biomass: There are strong causal relationships between fish abundance and the presence of tiger salamanders. A fairly strong negative correlation exists consistently in prairie wetlands between fish and salamanders (Batzner et al. 2006; Hentges and Stewart 2010). Biomass is straight-forward measure that is relatively easy to get from the same fyke nets that are set for fish while sampling a wetland. It also is a good metric to assess their density while factoring them into the WCI.
6. Aquatic Invertebrate Taxon Richness: Invertebrate assemblage characteristics are among the most valuable indicators of wetland condition and function (Rader et al. 2001; Genet and Olsen 2008). Invertebrate abundance and taxonomic diversity are clearly affected by habitat quality (Euliss et al. 1999; Rader et al. 2001). As consumers of vegetation, organic matter, and micro-organisms, and as prey for vertebrates, invertebrates also play critical roles in energy and nutrient flow in wetland food webs (Euliss et al. 1999; Woodcock et al. 2010). Strong associations exist between aquatic invertebrates and other wetland features. Generally, positive correlations exist with decreased turbidity, decreased fish biomass, and increased aquatic plant cover. Invertebrate Taxon Richness provides the strongest correlation to measure and can be measured on continuum basis conducive to the WCI.

METHODS FOR OBTAINING METRIC VALUES

This section of the protocol describes how to collect data and information for each of the six variables described above that can be then used as metric values in the Wetland Condition Index in order to determine the overall ecological condition of a wetland.

*** Please note that some of the variables will be lumped together in this section because the methods to collect them are completed together in the same manner with the same equipment.

*** It is important to collect the water quality related samples (turbidity and chloride) first in order to obtain a representative sample before disturbance occurs within the wetland sampling for the other variables.

Aquatic Invasive Species (AIS): Special care and planning should be taken by field sampling personnel to consult with the latest information available on AIS for steps and precautions necessary to minimize the spread of invasive species across wetlands. This is a serious topic to address with sampling gear and equipment.

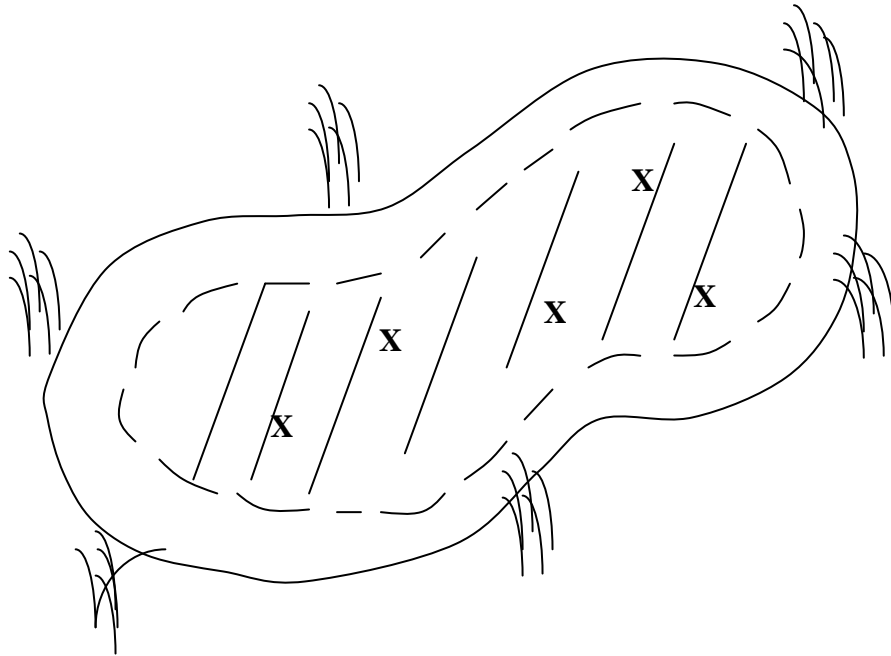
Basic Wetland Morphometry: These variables include wetland area, maximum depth, and mean depth. It is easiest and most efficient to quantify wetland area using a version of ArcGIS in advance of assessment field work of selected wetland sites. Maximum depth and mean depth can be obtained on site most likely during the aquatic plant survey work.

TURBIDITY

- **Gear and Equipment:** Two turbidimeters (in case one fails in the field) properly calibrated, clipboard with datasheet or electronic data device, chest waders, and canoe if needed.
- **Sampling Timeframe:** June 1 – August 15th

Samples should be collected in the open water zone if possible and at depths of 40 - 60 cm if they exist at the time of sampling. If this depth range does not exist, then use good judgment in sampling the main open water basin of the wetland. Turbidity should be measured on at least three different dates, at least 6 days apart, throughout the sampling timeframe listed above. If possible, it is best to spread the sampling dates as far apart from each other as possible, with at least one round of turbidity samples occurring in June to coincide with maximum aquatic plant growth. Measure turbidity three times in the middle of the water column. This can be done in one of two ways. The sampler can simply lower the turbidimeter vial upside down into the water column until they reach mid-depth and then invert the bottle upwards to fill it. The second method is to fill a clean water bottle in the same manner and then fill the turbidimeter vial with water from that bottle. Turbidity should be measured at five evenly spaced locations within a wetland, with locations distributed within the previously described range of sampling depth. Always be careful not to disturb the bottom sediment while collecting turbidity data.

OVERHEAD VIEW OF WETLAND WATER QUALITY SAMPLE POINTS

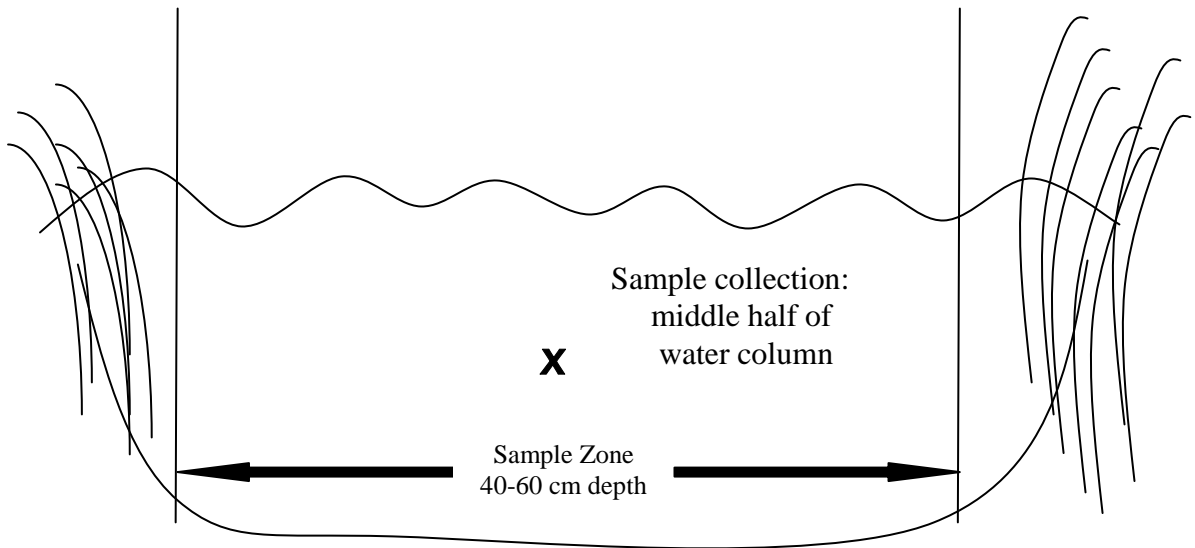


X – 5 Example of sample locations
(evenly spaced)



---- 40 – 60 cm sample zone depth

TURBIDITY SAMPLE



Cross Section of a Wetland

X – Turbidity sample location

CHLORIDE

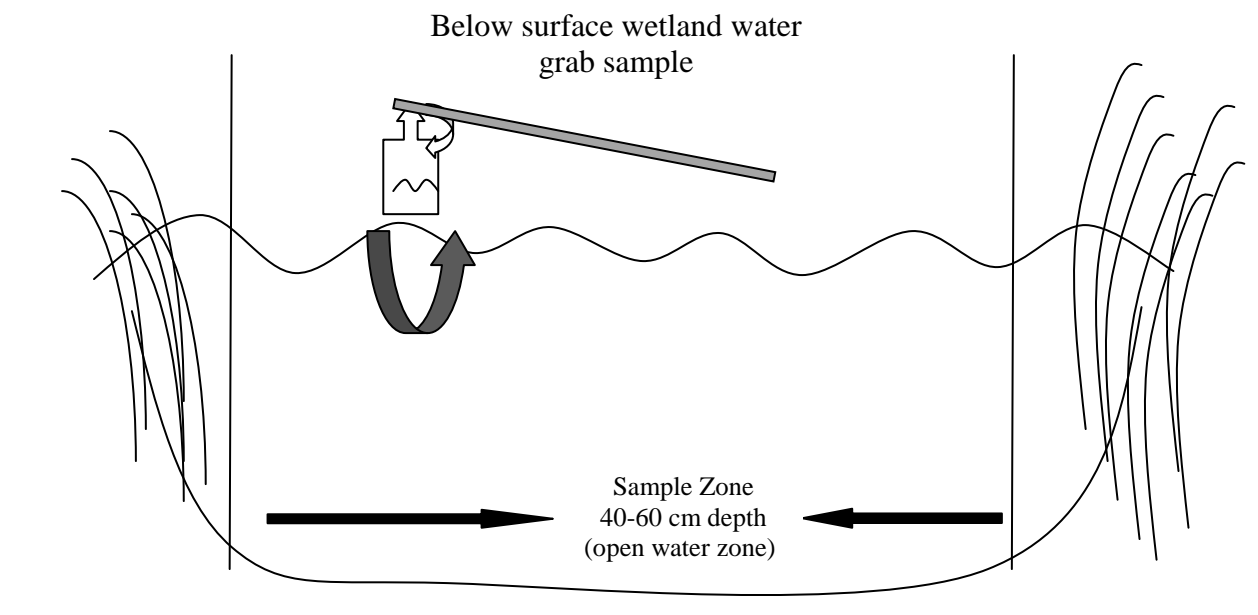
- **Gear and Equipment:** Chest waders, canoe, paddles, life jackets, sample bottles, water grab sample jug, cooler, permanent markers, and lab chain of custody forms.
- **Sampling Timeframe:** June 1 – August 15th

Wetland water samples collected to analyze for chloride can be collected 1 – 3 times throughout the sampling timeframe listed above. Three samples spread at least 6 days apart are ideal, but if time and/or budgets are limited, it can be sampled once per wetland. On each sampling date, collect one water grab sample from the middle of the water column in the middle of the wetland. A professionally accredited laboratory will need to analyze the water sample to get the chloride value. State of Iowa personnel will use the State Hygienic Laboratory (SHL) with the University of Iowa. The laboratory should provide water sample bottles for this sampling. The water grab sample can be collected in one of two ways generally:

1. Two field personnel access the middle of the wetland via canoe being careful not to disturb bottom sediment. Upon reaching the middle of the wetland, one person uses a thoroughly rinsed grab sample jug to fill water from the middle of the water column and then pour this water into the bottle provided by the laboratory.
2. For wetlands that are difficult to access, field personnel can carefully wade upwind to the middle of the wetland. Upon reaching the middle of the wetland, collect a water grab sample from the middle of the water column by reaching out into undisturbed water or use a long pole with the grab sample jug mounted to it to collect water. Then fill the lab bottle with water from this grab sample back at the shoreline.

After collecting the water samples for chloride analysis, field personnel should follow shipping guidelines provided by the laboratory to maintain quality control and assurance with each sample. Typically this will mean keeping the sample cool and out of direct sunlight in a cooler after collection, with bottles properly labeled with the collector's name, time, date, and site name on the bottle. Samples are then shipped or transported to the lab at the end of the field sampling portion of the day with chain of custody forms that track the samples from start to finish.

WETLAND CHLORIDE SAMPLING



FISH BIOMASS AND TIGER SALAMANDER BIOMASS

- **Gear and Equipment:** Chest waders, canoe, paddles, life jackets, scale, clipboard with datasheet or electronic data device, fish handling gloves, metal stakes to anchor nets, net anchors, bright colored flagging tape to mark net set locations, 3 standard fyke nets (15.2 m lead, 1.9-cm mesh, largest hoop opening = 0.6 x 1.2 m); 3 mini-fyke nets (4.0 m lead, 0.6-cm mesh, largest hoop opening = 0.6 x 1.2 m)
- **Sampling Timeframe:** June 15th – August 15th

Fish and salamander biomass only need to be sampled once during this timeframe.

Fyke nets should be oriented perpendicular to the shoreline and deployed in the open water zone at evenly spaced locations around the wetland much the same way they are set in lakes or ponds. In wetlands that have a very defined screen or wall of cattails between the shoreline and the open water zone, nets can be set up against the cattails such that fish cannot pass around the staked end of the net lead. Depending on water depth, field personnel can use a canoe or wade with chest waders to set the net properly, making sure the lead is on the wetland bottom all the way out to the hoop frame. Flagging tape can be wrapped around the net stakes on the shoreline so they are easy to spot the next day when checking nets. In large wetlands, it is also helpful to mark net site locations on a map in case different personnel check the nets. Nets should be checked and retrieved after a 24 hr period. Fish weight by species should be recorded. Tiger salamander weight (mainly in the mini-fyke nets) should also be recorded. It is recommended that field personnel place 1 -3 empty plastic jugs inside of the fyke nets so to prevent drowning of non-target animal species such as furbearers, turtles, or waterfowl. Care should be taken when checking the nets to avoid being bitten by non-target animals accidentally caught in the nets.

AQUATIC PLANT COVER (ABUNDANCE)

- **Gear and Equipment:** Chest waders, meter stick, data recording device or clipboard with datasheet, 1 m² plot (wood or metal stake with a ½ meter section of rope)
- **Sampling Timeframe:** July 1 – August 15th (preferably during July)

Aquatic plant cover only needs to be sampled once during this timeframe

Macroscopic aquatic plant assemblage consists of all free-floating and rooted floating-leaved, emergent, and submergent nonvascular and vascular taxa (Richardson and Vymazal 2001). Aquatic plants are surveyed using methods adapted from Johnston et al. (2009) and Kaeser and Kirkman (2009). Five parallel transects were established at evenly spaced locations, with each transect extending from shoreline to shoreline (defined by the uninterrupted presence of standing water). Each transect is divided into five sections of equal length, and one 1.0 m² sampling plot

randomly selected from each section. Visual observation and plant rake are used to estimate areal percent cover of plants in each plot. Methods for evaluating percent cover can also be further evaluated from Goldsmith and Harrison (1976). A meter stick is used to measure depth in the center of each plot. The meter stick can be modified to detect a soft bottom with a circular flat object of metal or wood attached at one end.

It is usually best if two field personnel work together as a pair to conduct this plant survey. It works best for the two field personnel to start at one end of the wetland at the beginning of a transect line and traverse the wetland transects in order across the wetland to the other end. One can carry the plot pole and walk the transects planting the plot pole at the appropriate spots to survey plant cover. The second person trails the first person closely and records data at each plot. Aquatic plant taxa are identified on-site, usually to genus, with species being identified when known. Plant cover for that wetland is estimated then from the 25 plots sampled within that wetland. Maximum and mean depth values are recorded from readings with the meter stick at each of these 25 plots as well.

AQUATIC INVERTEBRATE TAXON RICHNESS

- **Gear and Equipment:** Chest waders, 36-cm dia. Stovepipe sampler, white bug trays, forceps, sample containers, preservative, rose-bengal dye, 2 – 3 250 um mesh nets, permanent markers to label containers,
- **Sampling Timeframe:** June 15th – August 15th

Invertebrates only need to be sampled once during this timeframe.

Using a 36-cm diameter stovepipe sampler, a field crew of 2 – 3 people should sample at 5 evenly spaced locations (preferably in the 40 – 60 cm depth range) of the open water zone in a wetland (US EPA 2002; Hentges and Stewart 2010). The sampler should be placed firmly into the wetland sediment to a depth of 5 cm. The sampler needs to extend up through the water column with the top of it entirely out of the water in order to trap invertebrates from escaping out of it. Once the sampler is in place, sampling can occur. This is done by first removing pieces or clumps of aquatic plants by hand removing any invertebrates that are on the plant material and placing them into the sample container. It is helpful to have a white sample tray and forceps handy in which to place the plant material. Fill the tray with 1 – 3 cm of water to encourage invertebrates to swim off of the plant material to speed up the sorting process. Use a fine mesh (250-um) net to collect the top 2.5 cm of sediment and sweep it through the water column within the sampler until the organic matter is gone but the inverts remain. A sieve of the same mesh size can be used for sorting inverts from sediment as well. Lastly, sweep the mesh net through the water column within the sampler to catch remaining invertebrates. Do this until 10 consecutive sweeps produce no visible inverts. Preserve inverts in the container using 5% buffered formalin

(Rose Bengal dye optional to stain inverts). Formalin can be replaced with 70% ethanol after a 24 hr period. All five invertebrate subsamples can be combined for one composite sample for that wetland.

Each invertebrate sample is processed in a laboratory. Large inverts can be searched for first by placing sample contents into a pan, scanning the entire sample with unaided eye, and removing all inverts that are greater than or equal to 0.5 cm long (King and Richardson 2002; Hentges and Stewart 2010). Subsampling using a gridded tray marked with 27.5 cm² cells was used to sample for smaller inverts. One cell is randomly selected and a frame is placed around it. Invertebrates within that cell are plucked out and put into a petrie dish to view under a micro-scope at 10x. This is continued until at least two cells or at least 500 inverts are collected. Insects and mollusks were identified to the family level, while most other invertebrates are identified to order, class, or phylum. Invertebrate taxon richness is quantified as the total number of invertebrate taxa recorded from a wetland.

INSTRUCTIONS FOR SCORING METRICS

The six reliable wetland condition variables used in this protocol constitute the core metrics of a Wetland Condition Index for Iowa Prairie Pothole Region wetlands: fish biomass, chloride concentration, turbidity, plant cover (abundance), tiger salamander biomass, and invertebrate taxon richness. Each metric is scored by plotting its response across a causal gradient, and dividing the metric data into three sectors. For each of the six metrics, a 1, 3, 5 scoring system is used across a condition gradient. Therefore, wetlands in the lowest sector (poor condition) are assigned a score of “1”, wetlands in the middle sector (fair condition) are assigned a score of “3”, and wetlands in the upper sector (good condition) are assigned a score of “5”. A wetland condition index value is obtained for each wetland by summing scores for all six metrics.

Once the data has been collected using the methodologies described above, it is time to apply it by scoring the metrics. This section describes how to score the metrics using the data that has been collected from a wetland with this protocol.

Turbidity (NTU)

The unit of measure for turbidity is typically NTUs (Nephelometric Turbidity Units). The metric score for turbidity is fairly straight-forward to obtain for each wetlands. It is simply the Mean value of all your turbidity readings. To get this, a Mean value is calculated from all turbidity readings taken from the wetland during the three rounds of sampling to provide one representative metric value. Follow these steps to obtain the right Mean value:

Recall in the methods that 3 turbidity readings are taken from each of 5 sample spots in a wetland during each round of sampling. There are 3 rounds of sampling for turbidity.

Steps to calculate the average:

1. Calculate the Mean from the 3 NTU values recorded from each spot first. That will take the 15 (3 readings at 5 spots) values down to 5 values for one round of sampling in that wetland.
2. Then calculate the Mean for those 5 values. This will provide you with one Mean turbidity value for each round of turbidity sampling in that wetland. So if you conducted 3 rounds of turbidity sampling in a wetland, you will now have 3 values. One Mean value per round of sampling.
3. Then, simply calculate the Mean value from those 3 values to obtain 1 Mean value overall for that wetland. This will be the turbidity metric score to use in the Wetland Condition Index for that wetland.

Chloride (mg/L)

Recall in the methods that 1 sample is collected from the middle of the wetland to obtain a chloride concentration value. And that it is recommended 3 rounds of chloride are collected from a wetland during the field season. To obtain a metric score for chloride, simply calculate the Mean value from the 3 chloride values obtained during field sampling. This one Mean value of chloride is the chloride metric value to use in the Wetland Condition Index for that wetland.

Total Fish Biomass (kg)

Recall in the methods that only 1 round of sampling occurs for fish, but protocol recommends using 3 regular fyke nets, plus 3 mini-fyke nets in each wetland for a total of 6 nets per wetland. The total fish biomass is obtained simply by summing the mass of all fish species (large and small) captured in the nets. This will provide the metric value to use in the Wetland Condition Index for that wetland.

Tiger Salamander Biomass (g)

Same as with fish, all tiger salamanders captured in the 6 fyke nets per wetland should be weighed using grams as the unit of measure. The total weight is the metric value to use in the Wetland Condition Index.

Aquatic Plant Cover (Abundance) (%)

Recall from the methods that percent cover is estimated from the 25 plots sampled per wetland. To obtain the metric value for plant cover simply use the Mean of the percent cover estimate

from the 25 plots. The Mean percent cover is the metric value to use in the Wetland Condition Index.

Aquatic Invertebrate Taxon Richness (taxa/wetland)

Recall from the methods that the number of taxa are enumerated in the lab after field collection occurs from five locations in the wetland. Simply sum the number of different taxa counted from the lab sample. The number of taxa is the metric value to use in the Wetland Condition Index.

After summarizing the assessment data for a wetland in the manner explained above, the user will have six metric values to use in the Wetland Condition Index in which to obtain a final score in evaluating the ecological condition of a wetland.

So, the next step is to use the Wetland Condition Index score sheet.

INSTRUCTIONS FOR USING THE WCI SCORE SHEET

The WCI score sheet is on the next page and should be fairly easy to use.

Step 1: For each wetland, simply enter in the 6 metric values calculated from this protocol and described above into the score sheet column labeled ‘actual data derived value’: turbidity, chloride, tiger salamander biomass, total fish biomass, invertebrate taxon richness, and plant cover.

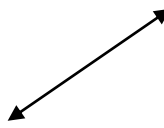
Step 2: Use the ‘Breakpoints for Each Metric’ column to decide which ‘Subsample Score’ to give each metric (1, 3, or 5). Enter this score into the column labeled ‘Metric Subsample Score’.

Step 3: Simply add up the ‘Total Score’ at the bottom of the column labeled ‘Metric Subsample Score’.

Step 4: Use the total score to determine which ‘Quality Range’ (poor, fair, or good) the wetland falls within. This is the final step in determining the current ecological condition (quality) a wetland is in.

WETLAND CONDITION INDEX SCORE SHEET

METRIC	Actual Data Derived Value	Breakpoints for each Metric	Metric Subsample Score (1, 3, 5)
-Turbidity (NTU) ----->		7.0 - 250 = 1 3.6 - 6.9 = 3 0 - 3.5 = 5	
-Chloride (mg/L) ----->		7.5 - 30.0 = 1 1.1 - 7.4 = 3 0 - 1 = 5	
-Tiger Salamander Biomass (g)--->		0 = 1 0.1 - 84.9 = 3 85 - 1600 = 5	
-Total Fish Biomass (kg)----->		2.0 - 150 = 1 .001 - 1.999 = 3 0 = 5	
-Invert Taxon Richness (# taxa/wetland) ----->		0 - 20 = 1 21 - 24 = 3 25 - 45 = 5	
-Plant Cover (%) ----->		0 - 87 = 1 87.1 - 96.9 = 3 97 - 100 = 5	
TOTAL SCORE			



<u>Quality Range</u>	
Poor	0 - 10
Fair	11 - 20
Good	21 - 30

INTERPRETATION OF A WETLAND CONDITION INDEX SCORE

The Wetland Condition Index is an effective tool for wetland assessment. Strong biotic and abiotic interactions exist within wetlands. So they are important to account for when assessing ecological condition in wetlands. The WCI helps to sort out which variables should be measured in order to collect the most representative data possible efficiently.

Interpretation of wetland scores derived from this protocol will help land managers to better understand the quality of the wetlands they manage. Because there six metrics used to assess these wetlands, it is possible to study which metrics need improvement if a wetland scores and 'poor' or 'fair'. Land managers can then take steps to improve the quality of the wetland. For example, if 'Total Fish Biomass' contributes heavily to a low score in the WCI for a particular wetland, a manager could then take steps to rid that wetland of rough fish by manipulating water levels. Over time, a manager can track the progress of a wetland's quality to gauge whether the wetland is improving or not using this assessment.

Wetlands in the PPR of Iowa may be extant (existing, never drained), restored, enhanced, or created. So, their functions and history varies. Therefore, management objectives may vary from wetland to wetland. People view wetlands in many different ways for many different reasons. Assessing the ecological condition of wetlands to determine their inherent quality as a wetland is the first step in assessing a wetland and the best approach ecologically to comprehensive wetland management. Specific management objectives may warrant further assessment to some wetlands, but this method provides a consistent way to track wetland quality over time.

Acknowledgements

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